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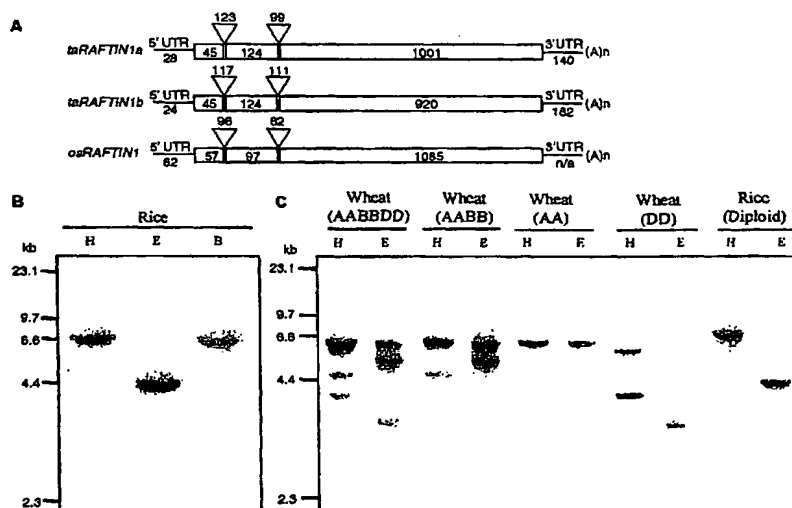
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(81) Designated States (national): AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW.

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[Continued on next page]

(54) Title: **RAFTIN GENE, PRODUCT, AND USE THEREOF**



(57) Abstract: The anther recruits a number of specific-genes to support microspore development. Here we report identification of a novel gene, RAFTIN1, from bread wheat and rice, encoding a protein containing a BURP domain hallmarked with 4 CH repeats at its C-terminus. This single copy gene (per haploid complement in cereals) is exclusively expressed in the tapetum during post-meiotic stages when the young microspore undergoes rapid expansion. RAFTIN1 is biosynthesized in the tapetum, transported into the Ubisch body, and further deposited onto the microspore exinewall. Transgenic rice, in which RAFTIN1 expression is down-regulated, shows normal growth and development but also shows male sterility. In the RAFTIN1-less anther, tapetal degeneration is retarded and pollen lacks contents. Thus, RAFTIN1 assembly in the Ubisch body and the microspore exinewall is required for microspore development, probably for regulation of metabolite transport from the tapetum to the microspore. Silencing or knocking-out of RAFTIN will find utility in breeding programs wherever male sterile lines are required.

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Saskatchewan S7N 4A5 (CA). **XIE, Wenshuang** [CA/US]; 5529 Colusa Avenue, Richmond, CA 94804-5227 (US). **DATLA, Raju** [CA/CA]; 527 Bayview Terrace, Saskatoon, Saskatchewan S7V 1B6 (CA).

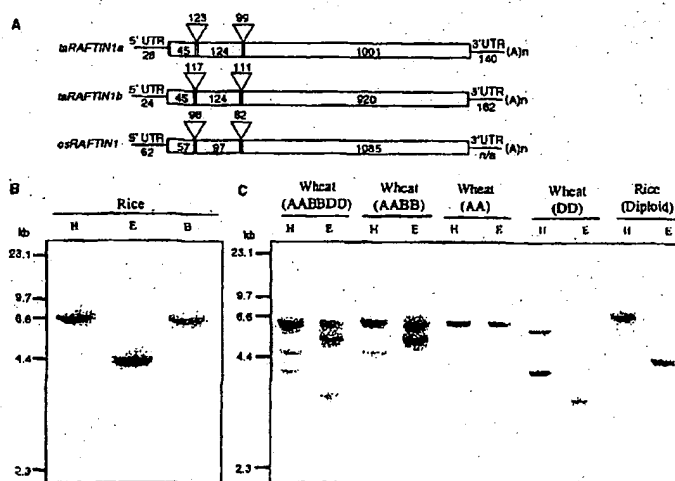
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[Continued on next page]

(54) Title: PROTEIN CONTAINING A BURP DOMAIN



(57) Abstract: The anther recruits a number of specific-genes to support microspore development. Here we report identification of a novel gene, RAFTIN1, from bread wheat and rice, encoding a protein containing a BURP domain hallmarked with 4 CH repeats at its C-terminus. This single copy gene (per haploid complement in cereals) is exclusively expressed in the tapetum during post-meiotic stages when the young microspore undergoes rapid expansion. RAFTIN1 is biosynthesized in the tapetum, transported into the Ubisch body, and further deposited onto the microspore exinewall. Transgenic rice, in which RAFTIN1 expression is down-regulated, shows normal growth and development but also shows male sterility. In the RAFTIN1-less anther, tapetal degeneration is retarded and pollen lacks contents. Thus, RAFTIN1 assembly in the Ubisch body and the microspore exinewall is required for microspore development, probably for regulation of metabolite transport from the tapetum to the microspore. Silencing or knocking-out of RAFTIN will find utility in breeding programs wherever male sterile lines are required.



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- before the expiration of the time limit for amending the claims and to be republished in the event of receipt of amendments

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INTERNATIONAL SEARCH REPORT

Application No
PCT/CA 03/01169

A. CLASSIFICATION OF SUBJECT MATTER
IPC 7 C07K14/415

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 7 C07K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

EPO-Internal, CHEM ABS Data, EMBL, Sequence Search, BIOSIS, WPI Data

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	BANZAI, TOSHIKI ET AL: "Molecular cloning and characterization of genes encoding BURP domain-containing protein in the mangrove, Bruguiera gymnorhiza" TREES (BERLIN, GERMANY) (2002), 16(2-3), 87-93, XP002259318 page 87, right-hand column, last paragraph - page 88, left-hand column, last paragraph	1-19
A	BATCHELOR, ANTHEA K. ET AL: "SCB1, a BURP-domain protein gene, from developing soybean seed coats" PLANTA (2002), 215(4), 523-532, XP002259319 abstract	1-19

☒ Further documents are listed in the continuation of box C.

☐ Patent family members are listed in annex.

* Special categories of cited documents:

- *A* document defining the general state of the art which is not considered to be of particular relevance
- *E* earlier document but published on or after the international filing date
- *L* document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)
- *O* document referring to an oral disclosure, use, exhibition or other means
- *P* document published prior to the international filing date but later than the priority date claimed

- *T* later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
- *X* document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
- *Y* document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art
- *G* document member of the same patent family

Date of the actual completion of the international search

27 October 2003

Date of mailing of the international search report

19.04.04

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INTERNATIONAL SEARCH REPORT

Int. Application No

PCT/CA 03/01169

*C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	HATTORI, J. ET AL: "A conserved BURP domain defines a novel group of plant proteins with unusual primary structures" MOLECULAR & GENERAL GENETICS (1998), 259(4), 424-428, XP002259320 the whole document	1-19
P, X	----- DATABASE EMBL 'Online! 22 November 2002 (2002-11-22), TINGEY S.V., MOORE G., GRIFFITHS S., POWELL W., WOLTERS P., DOLAN M., HAINEY C., MIAO G., CARAHER N., HANAFEY M.K.: "wpa1c.pk011.g9 wpa1c Triticum aestivum cDNA clone wpa1c.pk011.g9 5' end, mRNA sequence." XP002259321 retrieved from EBI Database accession no. CA596094 abstract	2-10
X	----- DATABASE EMBL 'Online! 5 June 2002 (2002-06-05), ZHANG H., WESCHKE W., MICHALEK W., STEIN N., GRANER A.: "HM01G17T HM Hordeum vulgare cDNA clone HM01G17 5-PRIME, mRNA sequence." XP002259322 retrieved from EBI Database accession no. BQ468492 abstract	2-10
X	----- DATABASE SWALL 'Online! 1 November 1999 (1999-11-01), ASAKI T., MATSUMOTO T., YAMAMOTO K.;: "Oryza sativa nipponbare(GA3) genomic DNA, chromosome 8, PAC" XP002259323 retrieved from EBI Database accession no. Q9XE32 abstract	2-10

INTERNATIONAL SEARCH REPORT

International application No.
PCT/CA 03/01169

Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This International Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☐ Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:

2. ☒ Claims Nos.: 1
because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful International Search can be carried out, specifically:
see FURTHER INFORMATION sheet PCT/ISA/210

3. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

see additional sheet

1. ☐ As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.

2. ☐ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.

3. ☐ As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:

4. ☒ No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:
partially 2-19

Remark on Protest

- ☐ The additional search fees were accompanied by the applicant's protest.
- ☐ No protest accompanied the payment of additional search fees.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

Continuation of Box I.2

Claims Nos.: 1

Present claim 1 lacks clarity contrary to Article 6 PCT as the internal arbitrary designation "RAFTIN" is unclear. Such an entity should be clearly and unambiguously characterized by reference to technical features such as SEQ ID NO. or by reference to the deposition number, which would unambiguously define the nucleotide sequence arbitrary designated "RAFTIN".

In fact, the claims contains so many options that a lack of clarity (and/or conciseness) within the meaning of Article 6 PCT and/or disclosure within the meaning of Article 5 PCT arises . The search has been carried out for those parts of the application which do appear to be clear (and/or concise), namely to the claimed subject-matter of present claims 2-19. Consequently no search has been performed for present claim 1.

The applicant's attention is drawn to the fact that claims relating to inventions in respect of which no international search report has been established need not be the subject of an international preliminary examination (Rule 66.1(e) PCT). The applicant is advised that the EPO policy when acting as an International Preliminary Examining Authority is normally not to carry out a preliminary examination on matter which has not been searched. This is the case irrespective of whether or not the claims are amended following receipt of the search report or during any Chapter II procedure. If the application proceeds into the regional phase before the EPO, the applicant is reminded that a search may be carried out during examination before the EPO (see EPO Guideline C-VI, 8.5), should the problems which led to the Article 17(2) declaration be overcome.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

This International Searching Authority found multiple (groups of) inventions in this international application, as follows:

1. claims: partially 2-19

Referr to the isolation and utilisation of taRAFTIN1a

2. claims: partially 2-19

Referr to the isolation and utilisation of taRAFTIN1b

3. claims: partially 2-19

Referr to the isolation and utilisation of taRAFTIN1d

4. claims: partially 2-19

Referr to the isolation and utilisation of osRAFTIN1
